

## IV Injection Recommendations for mice

For i.v. injections to mice, LASP recommends use of the lateral tail vein, while the mouse is enclosed within a commercial or custom mouse-restrainer. Dilation of the vein is accomplished by warming the tail by immersion in warm water or use of a heat lamp. Five minutes of supplemental heat may be necessary for optimal dilation. Monitor carefully to prevent the animal from being burned or overheated by the heat source.

Generally, the solution to be injected should be warmed to at least room temperature, be sterile, and have a pH between 4.5 and 8.0, with 7.4 being optimal.

Use of a ½ inch 26 or 27 ga needle (or smaller) with a 1 or 3 ml syringe is standard. Cells should be loaded with a larger (20+) gauge needle to minimize trauma to the cells. Changing needles also serves to keep the injection needle from becoming dulled if it is passed through a stoppered vial. The vein is best entered in the proximal 1/3 of the tail, and a successful injection is obvious to the user, based on lack of resistance as the plunger is depressed. Mastering of the injection technique takes practice, and training sessions may be scheduled by contacting the LASP veterinary staff.

The maximum volume that may be injected depends on numerous factors, including the mouse's size and background strain and properties of the compound, cells, or vehicle.. In most cases an injection volume of 0.2-0.5 ml can be safely given to an adult mouse. Most users try to limit the injection to 0.25 ml, a standard veterinary recommendation, but for cell injections, over-concentration may lead to embolism in lung capillaries and death of the recipient. The tendency to clump is cell-line-dependent. For example, a concentration of 40 million cells/ml of sarcoma cells may be fine, but larger, clumpy neuroblastoma cells must be given at no more than 10 million/ml, in a volume of 0.5 ml. For optimal volumes and numbers of cells, it is important to consult the experimental literature.

For hydrodynamic gene therapy, efficient transduction requires much higher injection volumes (YK Song et al, *Methods in Enzymology* 346:92-105, 2002). The usual rule is to give 10% of the body weight, so that a 20 g mouse would receive 2.0 ml of plasmid DNA over 3-5 seconds. B6 mice often have no reaction to such an injection, whereas Balb/c mice are more sensitive, displaying immobility with occasional respiratory compromise. If moderate to severe dyspnea is observed immediately following injection (uncommon), the animals should be euthanized or else supported with oxygen and supplemental heat. Most mice show only reduced activity levels. For routine hydrodynamic-based transfection, animals should be segregated and observed for the first hour following injection. If no problems are observed other than reduced activity, the mice may be returned to standard housing; normal behavior should be evident the next morning.